

contacting the analyte with said mixture, removing the unbound nucleounits by partitioning, amplifying the remaining nucleounits by PCR, conjugating the nucleounits to an indicator and then using the nucleounit indicator conjugate for detecting the presence of Prostate Specific Antigen by;

- (A) placing the reagent consisting essentially of an nucleounit indicator conjugate and a buffer in the reagent compartment of a chemistry autoanalyzer, aliquoting samples, calibrators, and controls into sample cups and placing them on the chemistry autoanalyzer;
- (B) and transferring aliquots of each sample, calibrator, and control into single discrete cuvettes mounted within the chemistry autoanalyzer; and
- (C) aliquoting the said reagent into each cuvette and mixing; and
- (D) incubating the reaction mixture; and
- (E) measuring and recording absorbance values of the reaction mixtures with the chemistry autoanalyzer's spectrophotometer at 340 nm to 800 nm at preprogrammed time intervals; and
- (F) comparing absorbance values of samples and controls to those of calibrators in the form of a standard curve thereby quantitating the prostate specific antigen present without the use of nucleic acid ligands.

Claim 13 (original): The method according to claim 12 wherein said nucleounit indicator conjugate can be selected from the group consisting of 4-Aminophenyl-beta-D-galactopyranoside, 3-indoxyl-beta-D-galactopyranoside (blue), 5-Bromo-4-chloro-3-indoxyl-beta-D-galactopyranoside (blue), 5-Bromo-3-indoxyl-beta-D-galactopyranoside (blue), 6-chloro-3-indoxyl-beta-D-galactopyranoside (salmon), 5-bromo-6-chloro-3-indoxyl-beta-D-galactopyranoside (Magenta- beta- D-Gal) 6-Fluoro-3-indoxyl-beta-D-galactopyranoside, 8-Hydroxyquinoline-beta-D-galactopyranoside, 5-Iodo-3-indoxyl-beta-D-galactopyranoside (purple), 2-Nitrophenyl-beta-D-galactopyranoside, 4-Nitrophenyl-beta-D-galactopyranoside, Naphthol AS-BI-beta-D-galactopyranoside, 2-

Naphthyl-beta-D-galactopyranoside (yellow), 4-Methylumbelliferyl-beta-D-glucuronic acid, galactosidase, beta-D-Galactosidase, Iodo-3-indoxyl-beta-D-galactopyranoside, alpha-L-Galactosidase, Iodo-3-indoxyl-alpha-L-galactopyranoside, beta-D-Cellobiosidase, 5-Bromo-4-chloro-3-indoxyl-beta-D-cellobioside, 5-Bromo-6-chloro-3-indoxyl-beta-D-cellobioside, 4-Nitrophenyl-beta-D-cellobioside, 1-Naphthyl-cellobioside, 4-Methylumbelliferyl-beta-D-cellobioside, glycosidases, galactosidase, cellobiosidase, Arabinosidase, Fucosidase, Galactosaminidase, Glucosaminidase, Glucosidase, Glucuronidase, Lactosidase, Maltosidase, Mannosidase, Xylosidase, arabinopyranoside, Fucopyranoside, Galactosaminide, Glucosaminide, Glucopyranoside, Glucuronic acid, Lactopyranoside, Maltopyranoside, Mannopyranoside, Xylopyranoside, 5-Bromo-4-chloro-3-indoxyl, 5-Bromo-6-chloro-3-indoxyl, 6-chloro-3-indoxyl, 5-Bromo-3-indoxyl, 5-Iodo-3-indoxyl, 3-indoxyl, 2-(6-Bromonaphthyl), 6-Fluoro-3-indoxyl 2-Nitrophenyl, 4-Nitrophenyl, 1-Naphthyl, Naphthyl AS-BI, 2-Nitrophenyl-N-acetyl, 4-Nitrophenyl-N-acetyl, 4-Methylumbelliferyl moieties, esterases, Carboxyl esterase, Cholesterol esterase, sulfatases, Aryl sulfatase, phosphatases, Alkaline phosphatase, Carboxyl esterase, 6-chloro-3-indoxyl butyrate, Aryl sulfatase, 5-bromo-4-chloro-3-indoxyl sulfate, alkaline phosphatase 2-naphthyl, dehydrogenase, oxidase, hydroxylase, oxidoreductase, dehydrogenases, hydroxylases, a-NAD, nicotinamide adenine dinucleotide, nicotinamide adenine dinucleotide 3'-phosphate, nicotinamide adenine dinucleotide phosphate, triphosphopyridine, nicotinamide 1-N1-ethenoadenine dinucleotide phosphate, nicotinamide hypoxanthine dinucleotide, nicotinamide hypoxanthine dinucleotide phosphate, nicotinamide mononucleotide, nicotinamide N1-propylsulfonate, nicotinamide ribose monophosphate, Formaldehyde dehydrogenase, Fructose dehydrogenase, Glucose-6-phosphate dehydrogenase, Glucose dehydrogenase, Glutamate dehydrogenase, Glycerol dehydrogenase, Glycerol-3-phosphate dehydrogenase, hydroxybutyrate dehydrogenase, Hydroxybenzoate hydroxylase, Lactate dehydrogenase, Leucine dehydrogenase, Malate dehydrogenase, Mannitol dehydrogenase, oxidases, Acyl-CoA oxidase, alcohol oxidase, ascorbate oxidase, Cholesterol oxidase, Choline oxidase, Glucose oxidase, Glycerophosphate oxidase, Xanthine oxidase, Uricase, peroxidase, pyrogallol, ABTS (2,2'-Azinobis(3-ethylbenzthiazoline) sulfonic acid), 3,3',5,5'-Tetramethylbenzidine, ortho-Dianisidine,

3,3'-Diaminibenzidine, AEC (3-Amino-9-ethyl carbazole), 2,5, dimethyl-2,5-dihydroperoxyhexane, Bis{4-[N-(3'-sulfo-n-propyl)-N-n-ethyl]amino-2,6-dimethylphenyl}methane (Bis-MAPS), N-Ethyl-N-(2-hydroxy-3-sulfopropyl)-3-methoxyaniline (ADOS), N-Ethyl-N-(3-sulfopropyl)-3-methoxyaniline (ADPS), N-Ethyl-N-(2-hydroxy-3-sulfopropyl)aniline (ALOS), N-Ethyl-N-(3-sulfopropyl)-3,5-dimethylaniline (MAPS), N-Ethyl-N-(2-hydroxy-3-sulfopropyl)-3-methylaniline (TOOS), N-Ethyl-N-(3-sulfopropyl)-3-methylaniline (TOPS), N-(3-sulfopropyl)aniline (HALPS), N-Ethyl-N-(2-hydroxy-3-sulfopropyl)-3,5-dimethoxyaniline (DAOS), N-Ethyl-N-(3-sulfopropyl)-3,5-dimethoxyaniline (DAPS), N-Ethyl-N-(3-sulfopropyl)aniline (ALPS), N-(2-hydroxy-3-sulfopropyl)-3,5-dimethoxyaniline (HDAOS), N-(3-sulfopropyl)-3,5-dimethoxyaniline (HDAPS), N-Ethyl-N-(2-hydroxy-3-sulfopropyl)-3,5-dimethylaniline (MAO), and N,N-Bis(4-sulfobutyl)-3,5-dimethylaniline (MADB), 3-Methyl-2-benzothiazolinonehydrazone, Dimerhylaniline, 4-aminoantipyrine, phenol, 2,4-Dichlorophenol, N,N-Diethyl-m-toluidine, p-Hydroxybenzene Sulfonate, N,N-Dimethylaniline, 3,5-Dichloro-2-Hydroxybenzenesulfonate, Sodium N-Ethyl-N-(3-Sulfopropyl)-m-Anisidine, N-Ethyl-N-(2-hydroxy-3-Sulfopropyl)-m-toluidine, NADPH oxidoreductase, NADPH, oxidoreductase, and NBT (nitro blue tetrazolium).

Claim 14 (original): The method according to claim 12 wherein said buffer can be selected from the group consisting of citrate, borate, phosphate, borax, sodium tetraborate decahydrate, sodium perchlorate, sodium chlorate, sodium carbonate, TRIS (Tris[hydroxymethyl]aminomethane), MES (2-[N-Morpholino]ethanesulfonic acid), BIS-TRIS (bis[2-Hydroxyethyl]iminotris[hydroxymethyl]methane; 2-bis[2-Hydroxyethyl]amino-2-[hydroxymethyl-1,3-propanediol]), ADA (N-[2-Acetamidol]-2-iminodiacetic acid; N-[Carbaoylmethyl]iminodiacetic acid), ACES (2-[(2-Amino-2-oxoethyl)amino]ethanesulfonic acid; N-[2-Acetamido]-2-aminoethanesulfonic acid), PIPES (Piperazine N-N'-bis[2-ethanesulfonic acid]); 1,4-Piperzinedethanesulfoic acid), MOPSO (3-[N-Morpholinol]-2-hydroxypropanesulfonic acid), BIS-TRIS PROPANE (1,3-bis[tris(Hydroxymethyl)methylamino]propane), BES (N,N-bis[2-Hydroxyethyl]-2-aminoethanesulfonic acid; 2-bis(2-Hydroxyethyl)amino]ethanesulfonic acid), MOPS (3-[N-Morpholino]propanesulfonic acid), TES (N-tris[Hydroxymethyl]methyl-2-

aminomethanesulfonic acid; 2[2-Hydroxy-1,1-bis(hydroxymethyl)-ethyl]amino)ethanesulfonic acid), HEPES (N-[2-Hydroxyethyl]piperazine-N'-[2-ethanesulfonic acid]), DIPSO (3-[N,N-bis(2-Hydroxyethyl)amino]-2-hydroxypropanesulfonic acid), TAPSO (3-[N-tris(Hydroxyethyl)methylamino]-2-hydroxypropanesulfonic acid), HEPPSO (N-[2-Hydroxyethyl]piperazine-N'-[2-Hydroxypropanesulfonic acid]), POPSO (Piperazine-N,N'-bis[2-hydroxypropanesulfonic acid]), EPPS (N-[2-Hydroxyethyl]piperazine-N'-[3-propanesulfonic acid]), TEA (triethanolamine), TRICINE (N-tris[Hydroxymethyl]methylglycine; N-[2-Hydroxy-1,1-bis(hydroxymethyl)ethyl]glycine), BICINE (N,N-bis[2-Hydroxyethyl]glycine), TAPS (N-tris[Hydroxymethyl]methyl-3-aminopropanesulfonic acid; ([2-Hydroxy-1,1-bis(hydroxymethyl)ethyl]amino)-1-propanesulfonic acid), AMPSO (3-[(1,1-Dimethyl-2-hydroxyethyl)amino]-2-hydroxypropanesulfonic acid), CHES (2-[N-Cyclohexylamino]ethanesulfonic acid), CAPSO (3-[Cyclohexylamino]-2-hydroxy-1-propanesulfonic acid), AMP 2-Amino-2-ethyl-1-propanol, CAPS (3-[cyclohexylamino]-1-propanesulfonic acid), hydrochloric acid, phosphoric acid, lactic acid, sulfuric acid, nitric acid, chromic acid, boric acid, perchloric acid, potassium hydrogen tartrate, potassium hydrogen phthalate, calcium hydroxide, phosphate, bicarbonate, sodium hydroxide, potassium hydroxide, oxalate or succinate.

Claim 15 (original): The method according to claim 12 wherein said analyte of interest can be selected from the group consisting of cocaine, opiates, gamma-hydroxybutyric acid, cannabinoids, benzodiazepines, acetaminophen, amikacin, aminocaproic acid, amitriptyline, amobarbital, amphetamine, bromide, caffeine, carbamazepine, carbenicillin, chloral hydrate, chloramphenicol, chlordiazepoxide, chlorpromazine, cimetidine, clonazepam, clonidine, clorazepate, cocaine, cocaine metabolites, ethanol, methanol, codeine, cyclosporine, desipramine, dexamethsone, diazepam, digoxin, diphenylhydantoin, disopyramide, doxepin, ephedrine, ethchlorvynol, ethosuximide, fenoprofen, flecainide, flurazepam, gentamicin, glutethimide, hydromorphone, ibuprofen, imipramine, isoniazid, kanamycin, lidocaine, lithium, lorazepam, lysergic acid, meperidine, meprobamate, methadone, methamphetamine, methaqualone, methotrexate,

methsuximide, methyldopa, methypylon, morphine, n-acetylprocainamide, netilmicin, nortriptyline, oxazepam, oxycodone, paraldehyde, paraquat, pentazocine, pentobarbital, phenacetin, phencyclidine, phenobarbital, phensuximide, phenylbutazone, phenylpropanolamine, phenytoin, primidone, procainamide, propoxyphene, propranolol, protriptyline, quinidine, salicylates, secobarbital, theophylline, thiocyanate, thiopental, thioridazine, tobramycin, tolbutamide, valproic acid, vancomycin, cholesterol, triglycerides, glucose, adrenocorticotrophic hormone, alanine, alanine aminotransferase, albumin, aldolase, aldosterone, amylase, amyloid-associated protein, androstenedione, angiotensin, antidiuretic hormone, antithrombin, antitrypsin, apolipoprotein, ascorbic acid, bile acids, bilirubin, c-peptide, calcitonin, calcium, cancer antigen 125, carboxyhemoglobin, carotene, catecholamines, cholic acid, cholyglycine, chromium, chymotrypsin, complement components, coproporphyrin, corticobinding globulin, corticosterone, cortisol, c-peptide, c-reactive protein, creatine, creatinine, creatine kinase, cyclic AMP, cystine, cysteine, dehydroepiandrosterone, dehydroepiandrosterone sulfate, deoxycholic acid, 11-deoxycorticosterone, 11-deoxycortisol, dihydrotestosterone, estradiol, estriol, estrogen, estrone, fecal fat, fatty acids, ferritin, fetoprotein, fibrinogen, folate, follicle stimulating hormone, thyroxine, triiodothyronine, fructose, fructosamine, galactose, gastric acid, gastrin, glucagons, glucose-6-phosphate, glutamine, glutamyltransferase (GGT), glutathione, hemoglobin, glycerol, glycine, glycolic acid, gold, gonadotropins, growth hormone, haptoglobin, high-density lipoproteins, hemopexin, homocystein, homocysteine, homogentisic acid, homovanillic acid, hydrogen sulfide, 17-hydroxycorticosteroids, 5-hydroxyindoleacetic acid, 17-hydroxyprogesterone, hydroxyproline, immunoglobins, insulin, iron, isocitrate dehydrogenase, isoleucine, 17-ketogenic steroids, ketone bodies, lactate, lactate dehydrogenase, lactose, LDL-cholesterol, lecithin, leucine, leukocyte, lipase, lipoproteins, lutropin, lysozyme, macroamylase, magnesium, melanin, metanephrine, methionine, metyrapone, microsomal antibodies, antibodies, molybdenum, mucopolysaccharide, myelin basic protein, myoglobin, methemoglobin, niacin, nickel, nitrite, nitrogen, nonprotein nitrogen, normetanephrine, blood, orosomucoid, oxalate, oxytocin, pancreatic polypeptide, pantothenic acid, parathyroid hormone, pentachlorophenol, pentoses, pepsinogen, phenols, phenolsulfonphthalein, phenylalanine, acid phosphatase, alkaline phosphatase,

phosphofructokinase, phospholipids, placental lactogen, plasminogen, porphobilinogen, prealbumin, pregnanediol, chorionic gonadotropin, pregnanetriol, pregnenolone, progesterone, porinsulin, properdin, prostaglandins, portoporphyrin, pseudocholinesterase, pyruvic acid, renin, reverse triiodothyromine, rheumatoid factor, riboflavin, secretin, selenium, serotonin, somatomedin c, sucrose, testosterone, tetrahydrocortisol, tetrahydrodeoxycortisol, thallium, thyroglobin, thyroid antibodies, thyroid stimulating hormone, thyroxine binding globulin, thyroxine, transcortin, transferrin, transketolase, transthyretin, thyrotropin-releasing hormone, triglycerides, triiodothyronine, tyrosine, urea, urea nitrogen, uric acid, uricase, urobilinogen, uroporphyrin, valine, vanillylmandelic acid, vasoactive intestinal polypeptide, human chorionic gonadotropin, mass creatinine kinase, vitamins, xylose, zinc, cyanide, formaldehyde, ethylene glycol, lead, mercury, xylene, human immunodeficiency virus (HIV), cytomegalovirus (CMV) IgG, cytomegalovirus (CMV) IgM, herpes simplex virus (types 1 and 2) IgG, rubella IgG, rubella, IgM, toxoplasma IgG, toxoplasma IgM, amebiasis, Epstein-barr early antigen, Epstein-barr EBNA IgG, Epstein-barr VCA IgG, Epstein-barr VCA IgM, helicobacter pylori-IgG, legionella IgG/IgM/IgA, mycoplasma IgG, mycoplasma IgM, varicella zoster virus (VZV), antinuclear antibodies (ANA), antineutrophil cytoplasmic antibodies (ANCA), anti-cardiolipin, anti-dsDNA, anti-Jo-1, anti-Scl-70, anti-Sm (Smith antigen), anti-Sm/RNP, anti-SS-A/Ro, anti-SS-B/La, extractable nuclear antigen (ENA), myeloperoxidase IgG, proteinase-3 IgG, or Rheumatoid Factor.